

Crystal deposition in hypophosphatasia: a reappraisal

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SUMMARY Six subjects (three female, three male; age range 38-85 years) with adult onset hypophosphatasia are described. Three presented typically with calcific periarthritides (due to apatite) in the absence of osteopenia; two had classical presentation with osteopenic fracture; and one was the asymptomatic father of one of the patients with calcific periarthritides. All three subjects over age 70 had isolated polyarticular chondrocalcinosis due to calcium pyrophosphate dihydrate crystal deposition; four of the six had spinal hyperostosis, extensive in two (Forestier's disease). The apparent paradoxical association of hypophosphatasia with calcific periarthritides and spinal hyperostosis is discussed in relation to the known effects of inorganic pyrophosphate on apatite crystal nucleation and growth.

Hypophosphatasia is a rare inherited disorder characterised by low serum levels of alkaline phosphatase, raised urinary phosphoethanolamine excretion, and increased serum and urinary concentrations of inorganic pyrophosphate.¹⁻⁴ Clinical presentation classically takes one of three forms according to age: failure to thrive with high mortality in infancy, a condition resembling childhood rickets, or multiple fractures due to severe osteopenia in adults.^{1,2} Disease severity is variable, especially in adult forms.^{1,2} An association with premature, polyarticular chondrocalcinosis due to calcium pyrophosphate dihydrate (CPPD) crystal deposition is well described.⁵⁻⁷

Figure 1 outlines the suggested mechanisms for the osteopenia and chondrocalcinosis characteristic of this disorder. The major biological source of inorganic pyrophosphate (PPi) derives from pyrophosphorolysis of nucleoside triphosphates and diphosphates during biosynthesis of most major cell constituents.⁸ Because alkaline phosphatase is the principal pyrophosphatase that converts PPi to orthophosphate, deficiency results in raised plasma and urinary concentrations of PPi.^{3,4,8,9} Inorganic pyrophosphate avidly binds to apatite crystals^{8,10-14} and, if unhydrolysed, is inhibitory to apatite crystal nucleation and growth,¹⁰⁻¹⁶ leading to poor mineralisation and predisposition to fracture. Persistent increase of PPi, in addition, increases the calcium ×

PPi ionic product, predisposing to enhanced CPPD crystal deposition in cartilage.

Paradoxical presentation with calcific periarthritides—that is, excess apatite, in three adults with biochemical hypophosphatasia but no osteopenia prompted us to re-examine these postulates. Findings in these cases are contrasted with those in two elderly patients with more classic hypophosphatasia presenting with severe osteopenia and multiple fractures, and the asymptomatic father of one of the patients with calcific periarthritides.

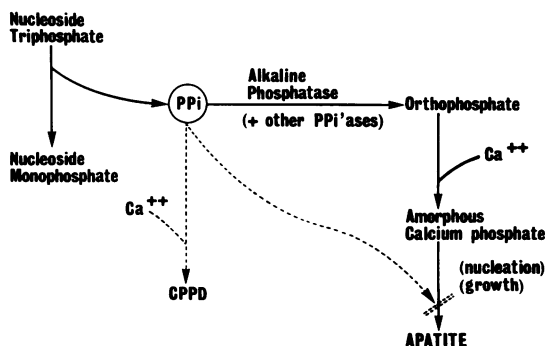


Fig. 1 Simplified scheme showing putative mechanisms linking hypophosphatasia with both osteopenia and calcium pyrophosphate dihydrate (CPPD) crystal deposition: lack of alkaline phosphatase results in increased inorganic pyrophosphate (PPi), which then inhibits apatite formation and growth and also predisposes to CPPD crystal formation.

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Patients and methods

Table 1 summarises the principal clinical, biochemical, and radiographic features of the six subjects. Each had consistently low serum alkaline phosphatase levels on repeat sampling: a metabolic screen, including serum urea, electrolytes, calcium, phosphate, magnesium, and thyroid function, failed to identify additional abnormality in any patient. Urinary phosphoethanolamine was estimated by high performance liquid chromatography after the method of Turnell and Cooper¹⁷ with quantification by standard amino acid analysis (expressed as mmol phosphoethanolamine/mmol creatinine) and was increased on at least one occasion in 4/5 subjects investigated. The urinary PPI (μmol)/creatinine (mmol) ratio was measured by a modification of the technique described by McGuire *et al*¹⁷ and was raised in all instances. Plain radiographs of hands, feet, knees, pelvis, and thoracic spine were obtained in all patients. The clinical histories are summarised in the following case reports.

CASE 1

A previously well 38 year old woman presented with acute onset of bilateral rotator cuff symptoms. Radiographs showed bilateral dense supraspinatus tendon calcification; needle aspiration of the left

shoulder deposit showed material positive on alizarin red staining,¹⁹ confirmed as hydroxyapatite by infrared spectroscopy. Her inflammatory symptoms slowly settled on treatment with naproxen and local steroid injection, but she persists with unaltered radiographic calcification and symptoms suggesting subacromial mechanical impingement.

CASE 2

A previously fit 47 year old woman presented with acute dactylitis of the left fourth toe, which resolved spontaneously over seven days. A radiograph (Fig. 2) showed ill defined soft tissue calcification at that site (absent on a radiograph repeated six months later); screening disclosed additional well defined calcific deposits close to the greater trochanters of both hips, periarticular calcification at one hip, and dense nummular calcification in one supraspinatus tendon. Calcification in the annuli of several discs was noted and confirmed by computed tomographic scan. In the thoracic spine hyperostotic changes typical of those reported in limited Forestier's disease²⁰ were present at multiple levels. She has had no further acute inflammatory episodes to date.

CASE 3

A previously fit 52 year old visiting Polish man presented with acute pain, swelling, and erythema

Table 1 Clinical, biochemical, and radiographic features of the subjects

	Patient No					
	1	2	3	4	5	6
Sex, age	F, 38	F, 47	M, 52	M, 72	F, 78	M, 85
Clinical features						
Calcific periarthritis	+	+	+		+	
Fractures				+		
Acute pseudogout				+		
Biochemical features						
Mean (n=2-5) serum alkaline phosphatase (U/l)*	73	60	60	48	31	40
Max urine PEA† (mmol)/creatinine (mmol)*	Not detected	0.046	No urine obtained	0.047	0.068	0.018
Max urine PPI‡ (μmol)/creatinine (mmol)*	14	60		56	79	11
Radiographic features						
Periarticular calcification	+	+	+			
Chondrocalcinosis				+	+	+
Spinal hyperostosis/Forestier's		+	+	+		++
Osteopenia				+	+	

*Normal values: serum alkaline phosphatase 80-280 U/l; urine phosphoethanolamine (mmol)/creatinine (mmol) ratio <0.010; urine PPI (μmol)/creatinine (mmol) ratio 0-5.

†PEA=phosphoethanolamine; PPI=inorganic pyrophosphate.

‡Plus diffuse idiopathic skeletal hyperostosis (DISH).



Fig. 2 *Left fourth toe radiograph (case 2) showing ill defined calcification, absent on a repeat radiograph six months later.*

around the left wrist. The radiograph showed discrete nummular calcification overlying the left carpus (Fig. 3), and needle aspiration showed calcific material with a spectrum on infrared spectroscopy consistent with carbonated hydroxyapatite. His acute inflammation settled on treatment with non-steroidal anti-inflammatory drugs alone, and a repeat wrist radiograph at 10 days, just before his return to Poland, showed considerable dispersal of the calcification (Fig. 3). Other radiographs showed normal mineralisation with no evidence of peripheral joint chondrocalcinosis or arthropathy. In the thoracic spine, however, marked calcification within the nucleus pulposus was present together with hypertrophic bone changes at multiple levels in a pattern consistent with limited Forestier's disease (Fig. 4).

CASE 4

A 72 year old man gave a 16 year history of multiple stress fractures and recurrent acute attacks of knee synovitis. The radiographic survey showed generalised osteopenia, multiple healed fractures, florid changes of Forestier's disease together with concave end plate depression in the thoracic spine, and polyarticular chondrocalcinosis of fibrocartilage and hyaline cartilage (knees, wrist triangular ligaments,



Fig. 3 *Lateral left wrist radiograph (case 3) showing discrete calcification (left), which had largely dispersed on the repeat film 10 days later (right).*

symphysis pubis) with no arthropathic change. Iliac crest bone biopsy showed widened osteoid seams; aspiration of both knees disclosed numerous crystals with morphology and polarised light characteristics of CPPD (no definitive crystal identification performed).

CASE 5

A 78 year old woman gave a 28 year history of multiple stress fractures. An iliac crest bone biopsy had shown features of osteomalacia, but her condition had proved resistant to vitamin D. Her radiographs showed severe osteopenia with old fractures and polyarticular chondrocalcinosis of hyaline and fibrocartilage (knees, wrist) but no changes of arthropathy. Synovial fluid from one asymptomatic knee showed CPPD crystals (confirmed by infrared spectroscopy).

CASE 6

This 85 year old man was the father of patient 3. He

had experienced no locomotor symptoms but on investigation was found to have hypophosphatasia, extensive radiographic chondrocalcinosis (knees only) without arthropathy, normal mineralisation, and florid changes of Forestier's disease (Fig. 4): in addition, he showed ossification at the site of several entheses (ischial spines, acetabular labra, patella and Achilles tendon insertions, plantar fascia insertions) typical of diffuse idiopathic skeletal hyperostosis (DISH). Examination of small volumes of synovial fluid aspirated from both asymptomatic knees showed positively birefringent rhomboid crystals, confirmed as CPPD by Fourier transform infrared spectroscopy.

Discussion

Hypophosphatasia in these cases was diagnosed by consistently low serum levels of alkaline phosphatase (all cases), increased urinary phosphoethanolamine excretion (on at least one occasion in four of

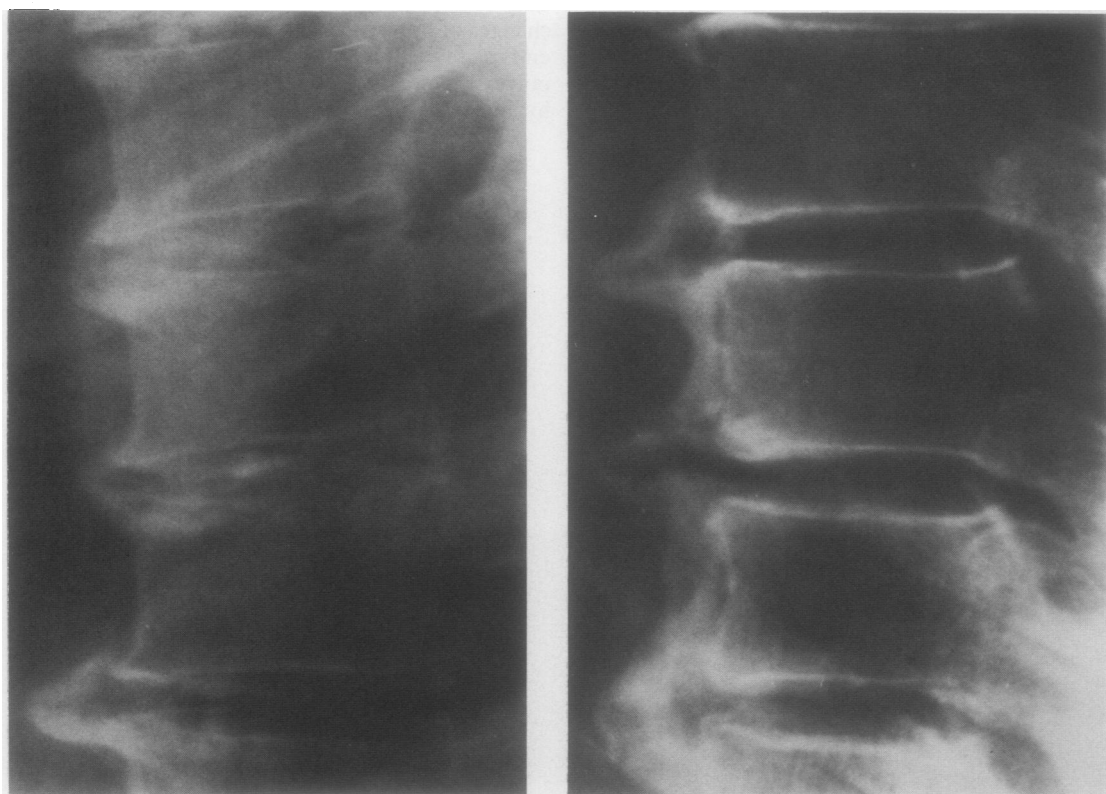


Fig. 4 Lateral thoracic spine radiographs of case 3 (left) and case 6 (right). In the younger patient (case 3) there is intervertebral disc calcification (nucleus pulposus), normal intervertebral height, and anterolateral hypertrophic changes at multiple levels; in the older subject (case 6) there is normal intervertebral height and similar but more pronounced new bone formation typical of Forestier's disease.

five subjects tested), and raised urinary PPi (in five cases tested). Interestingly, phosphoethanolamine excretion, usually regarded as mandatory for diagnosing hypophosphatasia, was variably positive in our subjects,²¹ whereas urinary PPi excretion was consistently increased.³ Although two patients had classic adult presentation with osteopenic fracture,^{1,2} three had calcific peri-arthritis, an association noted in just two previous reports.^{22,23} Although CPPD crystals may deposit in extracartilaginous sites,²⁴⁻²⁷ diagnosis of apatite associated peri-arthritis was by clinical and radiographic characteristics — for example, nummular not linear tendon calcification,²⁴ with definitive crystal identification in two cases. Of further interest was the occurrence of early or established features of spinal hyperostosis (Forestier's disease) in four subjects. Spinal new bone formation in hypophosphatasia is well recognised, but a pattern typical of Forestier's disease has been emphasised in only one report.²²

Although severe osteopenia is the expected consequence of hypophosphatasia, calcific peri-arthritis with normal bone density in three of our patients suggests that in certain situations low alkaline phosphatase levels may associate with stimulation rather than inhibition of apatite formation or growth. In vitro study of calcification mechanisms in epiphyseal growth cartilage^{10,12,13,15,16} shows that at high concentration PPi is a potent inhibitor of apatite: adsorption of PPi onto the amorphous calcium/phosphate complex prevents transformation to crystalline hydroxyapatite, and adsorption onto apatite crystals inhibits aggregation and growth.^{8,10-16} At lower concentrations, however, PPi promotes apatite crystal transformation and growth.^{10,12,15,16} Inorganic pyrophosphate is conspicuously present in mineralising tissues and is thought by its dual effect to be a physiological regulator of matrix vesicle calcification.¹⁵ Although osteoarticular concentrations were not assessed, it is conceivable that tissue PPi concentrations were in the range to stimulate apatite nucleation and growth in the three patients with calcific peri-arthritis (ectopic apatite), normal bone density, and modestly low serum alkaline phosphatase (mean (SD) 64.3 (7.5) U/l), whereas in those with osteopenia (insufficient apatite) and lower alkaline phosphatase levels (39.5 (12) U/l) tissue PPi was sufficiently raised to be inhibitory.

It is uncertain whether the three patients with calcific peri-arthritis represent a discrete mild subset of hypophosphatasia or the early development phase of classic osteopenic disease. Although peri-articular calcification is often asymptomatic, and conceivably may previously have been present in the older subjects, lack of osteopenia in the father (case

6) of patient 2 supports a less severe variety. Serial alkaline phosphatase levels show variation in patients with hypophosphatasia, however, and it is possible that PPi concentrations in individuals may fluctuate from inhibitory to stimulatory. If spinal hyperostosis also reflects PPi related stimulation of mineralisation, such temporal fluctuation may explain concurrence of osteopenia and Forestier's disease in some cases. Spinal changes in two of our younger patients were typical of Forestier's disease,^{28,29} but were not sufficiently extensive to fulfil Resnick's criteria.³⁰ The development of Forestier's disease remains unclear, but it seems likely that such changes represent early features of more florid, typical disease²⁰ seen in the two older subjects. Although four had axial hyperostosis, only one (case 6) had extraspinal new bone at the site of several entheses typical of diffuse idiopathic skeletal hyperostosis,^{29,30} again supporting a milder variant of hypophosphatasia in those without osteopenia.

In addition to its effects on apatite, PPi itself may be incorporated as crystalline CPPD ($\text{Ca}_2\text{P}_2\text{O}_7 \cdot 2\text{H}_2\text{O}$). Unlike apatite, CPPD crystal formation is mainly restricted to collagenous locomotor tissues (fibrocartilage and hyaline cartilage, tendon, capsule), and is particularly influenced by unidentified tissue factors that accompany aging.^{8,31,32} All three patients over age 70 had chondrocalcinosis with synovial fluid CPPD crystal confirmation: this could reflect either the independent predisposing effect of aging, or the duration and extent of increased tissue PPi. Interestingly, only one had joint symptoms (acute pseudogout), and none had radiographic features of arthropathy. A similar clinicoradiographic pattern (asymptomatic or acute attacks, with isolated radiographic chondrocalcinosis) occurs in three other metabolic diseases predisposing to CPPD crystal deposition — namely, hyperparathyroidism,^{33,34} hypomagnesaemia,^{35,36} and Wilson's disease.³⁷ Such similarity supports predisposition by effects on PPi metabolism — for example, inhibition of alkaline phosphatase by calcium, cupric, and ferrous ions³⁸; reduction in magnesium as co-factor for alkaline phosphatase³⁹; stimulation of adenylate cyclase by parathyroid hormone.^{8,39} By contrast, in haemochromatosis (another predisposing metabolic disease), although isolated chondrocalcinosis may occur, chronic symptoms with arthropathic change (cartilage/bone attrition, prominent cysts; distinctive distribution; often no overt synovitis) are more usual,^{40,41} suggesting that other non-PPi related mechanisms may be involved in predisposition to chondrocalcinosis and arthropathy. Further study of diseases showing isolated metabolic defects may give insight into both physiological and

pathological mechanisms of calcium crystal deposition.

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References

- Rasmussen H, Bartter F C. Hypophosphatasia. In: Stanbury J B, Wyngaarden J B, Frederickson D S, eds. *Metabolic basis of inherited disease*. 4th ed. New York: McGraw-Hill, 1978: 1340.
- Whyte M P, Murphy W A, Fallon M D. Adult hypophosphatasia with chondrocalcinosis and arthropathy. *Am J Med* 1982; **72**: 631-41.
- Russell R G G. Excretion of inorganic pyrophosphate in hypophosphatasia. *Lancet* 1965; **ii**: 461-4.
- Russell R G G, Bisaz S, Donath A, Morgan D B, Fleisch H. Inorganic pyrophosphate in plasma in normal persons and in patients with hypophosphatasia, osteogenesis imperfecta and other diseases of bone. *J Clin Invest* 1971; **50**: 961-9.
- O'Duffy J D. Hypophosphatasia associated with calcium pyrophosphate dihydrate deposits in cartilage. *Arthritis Rheum* 1970; **13**: 381-8.
- Eade A W T, Swannell A J, Williamson N. Pyrophosphate arthropathy in hypophosphatasia. *Ann Rheum Dis* 1981; **40**: 164-70.
- Pinquier J L, Awada H, Delfieu F, Amor B. Chondrocalcinose isolée relevant une hypophosphatasie de l'adulte. *Rev Rhum Mal Osteoartic* 1987; **54**: 501-4.
- Russell R G G. Metabolism of inorganic pyrophosphate. *Arthritis Rheum* 1976; **19**: 465-78.
- Fernley H N, Walker P G. Studies on alkaline phosphatase. Inhibition by phosphate derivatives and the substrate specificity. *Biochem J* 1967; **104**: 1011-8.
- Fleisch H, Strauman F, Schenk R, Bisaz S, Allgower M. Effect of condensed phosphates on calcification of chick embryo femurs in tissue culture. *Am J Physiol* 1966; **211**: 821-5.
- Fleisch H, Bisaz S. Isolation from urine of pyrophosphate, a calcification inhibitor. *Am J Physiol* 1962; **203**: 671-5.
- Seigel S A, Hummel C F, Carty R P. The role of nucleoside triphosphate pyrophosphohydrolase in in vitro nucleoside triphosphate-dependent matrix vesicle calcification. *J Biol Chem* 1983; **258**: 8601-7.
- Russell R G G, Fleisch H. Pyrophosphate and diphosphonates in skeletal metabolism. *Clin Orthop* 1975; **108**: 241-63.
- Hansen N M, Felix R, Bisaz S, Fleisch H. Aggregation of hydroxyapatite crystals. *Biochem Biophys Acta* 1976; **451**: 549-59.
- Hsu H H T, Anderson H C. The deposition of calcium pyrophosphate and phosphate by matrix vesicle isolated from fetal bovine epiphyseal cartilage. *Calcif Tissue Int* 1984; **36**: 615-21.
- Einhorn T A, Gordon S L, Siegal S A, Hummel C F, Avitable M J, Carty R P. Matrix vesicle enzymes in human osteoarthritis. *J Orthop Res* 1985; **3**: 160-9.
- Turnell D C, Cooper J D H. Rapid assay for amino acids in serum or urine by pre-column derivatization and reversed phase liquid chromatography. *Clin Chem* 1982; **28**: 527-31.
- McGuire M B, Colman C H, Baghat N, Russell R G G. Radiometric measurement of pyrophosphate in cell cultures. *Biochem Soc Trans* 1980; **8**: 529-30.
- Paul H, Reginato A J, Schumacher H R. Alizarin red S staining as a screening test to detect calcium compounds in synovial fluid. *Arthritis Rheum* 1983; **26**: 191-200.
- Fornasier V L, Littlejohn G, Urowitz M B, Keystone E C, Smythe H A. Spinal enthesal new bone formation: the early changes of spinal diffuse idiopathic skeletal hyperostosis. *J Rheumatol* 1983; **10**: 939-47.
- Pimstone B, Eisenberg E, Silverman S. Hypophosphatasia: genetic and dental studies. *Ann Intern Med* 1966; **65**: 722-9.
- Macfarlane J D, Kroon H M, Cats A. Ectopic calcification in hypophosphatasia. *Eur J Radiol* 1986; **6**: 228-30.
- Caspi D, Rosenbach T O, Yaron M, McCarty D J, Graff E. Periarthritis associated with basic calcium phosphate crystal deposition and low levels of serum alkaline phosphatase-report of three cases from one family. *J Rheumatol* 1988; **15**: 823-7.
- Gerster J C, Baud C A, Lagier R, Boussina I, Fallet G H. Tendon calcifications in chondrocalcinosis. A clinical, radiologic, histologic, and crystallographic study. *Arthritis Rheum* 1977; **20**: 717-22.
- Gerster J C, Lagier R, Boivin G. Olecranon bursitis related to calcium pyrophosphate dihydrate crystal deposition disease. A clinical and pathologic study. *Arthritis Rheum* 1982; **25**: 989-96.
- Leisen C C, Austad E D, Bluhm G B, Sigler J W. The tophus in calcium pyrophosphate deposition disease. *JAMA* 1980; **244**: 1711-2.
- Schumacher H R, Bonner H, Thompson J J, Kester W L, Benner J J. Tumor-like soft tissue swelling of the distal phalanx due to calcium pyrophosphate dihydrate crystal deposition. *Arthritis Rheum* 1984; **27**: 1428-32.
- Forestier J, Lagier R. Ankylosing hyperostosis of the spine. *Clin Orthop* 1971; **74**: 65-83.
- Resnick D, Shapiro R F, Wicner K B, Niwayama G, Utsinger P D, Shaul S R. Diffuse idiopathic skeletal hyperostosis (DISH) (ankylosing hyperostosis of Forestier and Rotes-Querol). *Semin Arthritis Rheum* 1987; **7**: 153-87.
- Resnick D, Shaul S, Robins J. Diffuse idiopathic skeletal hyperostosis: extraspinal manifestations of Forestier's disease. *Radiology* 1975; **115**: 513-24.
- Buckwalter J A. Proteoglycan structure in calcifying cartilage. *Clin Orthop* 1983; **172**: 207-32.
- Doherty M, Dieppe P A. Crystal deposition disease in the elderly. *Clin Rheum Dis* 1986; **12**: 97-116.
- Dodds W J, Steinbach H L. Primary hyperparathyroidism and articular cartilage calcification. *American Journal of Roentgenology, Radium Therapy and Nuclear Medicine* 1968; **104**: 884-92.
- Pritchard M H, Jessop J D. Chondrocalcinosis in primary hyperparathyroidism. Influence of age, metabolic bone disease, and parathyroidectomy. *Ann Rheum Dis* 1977; **36**: 146-51.
- Runeberg L, Collan Y, Jokinen E J, Lahdevirta J, Aro A. Hypomagnesaemia due to renal disease of unknown etiology. *Am J Med* 1975; **59**: 873-81.
- Milazzo S C, Ahern M J, Cleland L G, Henderson D R F. Calcium pyrophosphate dihydrate deposition disease and familial hypomagnesaemia. *J Rheumatol* 1981; **8**: 767-71.
- Feller E R, Schumacher H R. Osteoarticular changes in Wilson's disease. *Arthritis Rheum* 1972; **15**: 259-66.
- McCarty D J, Pepe P F, Solomon S D, Cobb J. Inhibition of human erythrocyte pyrophosphate activity by calcium, cupric and ferrous ions. *Arthritis Rheum* 1970; **13**: 336.
- Caswell A, Guillard-Cumming D F, Hearn P R, McGuire M K B, Russell R G G. Pathogenesis of chondrocalcinosis and pseudogout. Metabolism of inorganic pyrophosphate and production of calcium pyrophosphate dihydrate crystals. *Ann Rheum Dis* 1983; **42** (suppl): 27-37.
- Atkins C J, McIvor J, Smith P M, Hamilton E, Williams R. Chondrocalcinosis and arthropathy: studies in haemochromatosis and in idiopathic chondrocalcinosis. *Q J Med* 1970; **39**: 71-82.
- Hamilton E B D, Bomford A B, Laws J W, Williams R. The natural history of arthritis in idiopathic haemochromatosis: progression of the clinical and radiological features over ten years. *Q J Med* 1981; **199**: 321-9.